Registry No. **2a**, 86994-33-4; **2b**, 86994-34-5; **3a** (isomer 1), 86994-35-6; **3a** (isomer 2), 87038-36-6; **3b** (isomer 1), 86994-36-7; **3b** (isomer 2), 87038-37-7; **4**, 86994-37-8; **5**, 86994-38-9; **6**, 86994-39-0; **7**, 17692-20-5; **8**, 86994-40-3; 2′-hydroxyacetophenone, 118-93-4; 2′-methoxyacetophenone, 579-74-8; *tert*-butyl acetate, 540-88-5; acetylcyclohexane, 823-76-7; ethyl α-bromoacetate, 105-36-2; ethyl 3-hydroxy-3-cyclohexylbutyrate, 28811-84-9; 4-methylcoumarine, 607-71-6; cholesterol, 57-88-5.

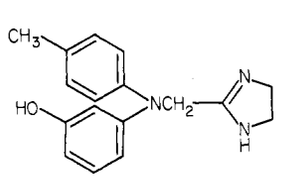
α-Adrenergic Activities of Some Substituted 2-(Aminomethyl)imidazolines

Walfred S. Saari,*† Wasyl Halczenko,† William C. Randall,† and Victor J. Lotti*‡

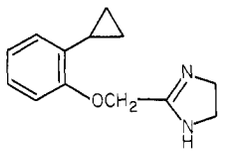
Merck Sharp & Dohme Research Laboratories, West Point, Pennsylvania 19486, and Merck Institute for Therapeutic Research, West Point, Pennsylvania 19486. *Received April 11, 1983*

A series of 2-(aminomethyl)imidazolines related to the α-adrenergic antagonist phentolamine was prepared and evaluated for α-adrenergic agonist and antagonist activities in the isolated, field-stimulated rat vas deferens. Affinities for α-adrenergic receptors were determined by displacement of [^{3}H]clonidine and [^{3}H]prazosin from membrane binding sites of calf cerebral cortex. This series provided a variety of α-adrenergic profiles, with some of the (aminomethyl)imidazolines being nonselective α_1- and α_2-adrenergic antagonists like phentolamine, while others were either nonselective α_1- and α_2-agonists or mixed α_1-agonists/α_2-antagonists.

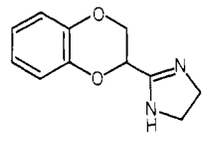
The α-adrenoceptor blocking agent phentolamine (**1**) is



1



2



3

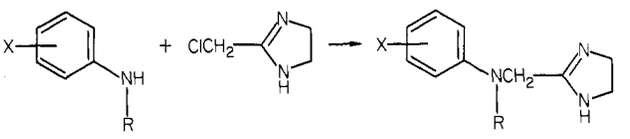
recognized to be a potent, but nonselective, competitive antagonist at both α_1- and α_2-adrenoceptor sites.[1,2] However, other imidazoline derivatives have recently been found to possess different profiles of adrenergic activities. For example, cirazoline (**2**) is described as having α_1-agonist and α_2-antagonist activities,[3] while RX 781094 (**3**) is reported to be a potent and selective α_2-antagonist with weaker α_1-antagonism properties.[4]

It was therefore of interest to characterize those molecular features of phentolamine responsible for α_1- and α_2-adrenoceptor antagonism and to use this information for the synthesis of more selective adrenergic blocking agents.

Chemistry. All of the imidazoline derivatives of Table I were synthesized by reaction of the appropriate arylamine with 2-(chloromethyl)imidazoline (Scheme I).[5,6] The required aniline derivatives have been reported in the literature and are readily available. However, in the case of the *N*-cyclohexyl-*N*-(4-tolyl) analogue **4f**, a two-step reductive alkylation of 4-methylaniline with cyclohexanone proved to be a more convenient preparation of the amine than the previously published procedure.

Testing Methods. Relative affinities of the imidazolines of Table I for central α-adrenergic binding sites were determined by measurement of radioligand displacement from membrane binding sites of calf cerebral cortex. Displacement of [^{3}H]clonidine was used as a measure of interaction with α_2-adrenoceptor binding sites, while

Scheme I



4

[^{3}H]prazosin displacement served as an assay for α_1-adrenoceptor affinity.

α-Adrenergic agonist–antagonist profiles for these compounds were determined in the rat, isolated, field-stimulated vas deferens according to protocols described by Lotti et al.[7] In this tissue, presynaptic (α_2) adrenergic agonists characteristically inhibit stimulation-induced contractions, while postsynaptic (α_1) agonists enhance contractions. The α_1- and α_2-adrenergic agonist activities of the test compounds were verified by the ability of prazosin and rauwolscine to completely reverse the contractile enhancement or inhibition, respectively, produced by the test compounds. Antagonistic activities of the imidazolines upon α_1- and α_2-adrenoceptors were determined by blockade of the selective α_1- and α_2-adrenergic agonists methoxamine and clonidine, respectively.

Results and Discussion

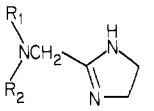
Inspection of the rat vas deferens results summarized in Table II shows that phenotolamine remains the most potent α_1/α_2-adrenoceptor antagonist in this series. Replacement of the phenolic hydroxyl group of phentolamine by hydrogen gives **4a**, which, although slightly less potent than phentolamine, still functions as a nonselective α-ad-

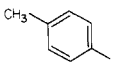
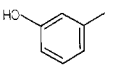
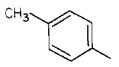
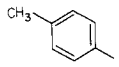
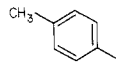
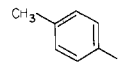
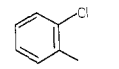
† Merck Sharp & Dohme Research Laboratories.
‡ Merck Institute for Therapeutic Research.

(1) Langer, S. Z. *Br. J. Pharmacol.* **1977**, *60*, 481.
(2) Starke, K.; Docherty, J. R. *J. Cardiovasc. Pharmacol.* **1980**, *2*(Suppl 3), S269.
(3) Hannah, J. A. M.; Hamilton, C. A.; Reid, J. L. *Br. J. Pharmacol.* **1982**, *75*, 141P. Cavero, I.; Langer, S. Z. *Ibid.* **1982**, *75*, 153P.
(4) Chapleo, C. B.; Doxey, J. C.; Myers, P. L.; Roach, A. G. *Br. J. Pharmacol.* **1981**, *74*, 842P. Chapleo, C. B.; Myers, P. L.; Butler, R. C. M.; Doxey, J. C.; Roach, A. G.; Smith, C. F. C. *J. Med. Chem.* **1983**, *26*, 823.
(5) Urech, E.; Marxer, A.; Miescher, K. *Helv. Chim. Acta* **1950**, *33*, 1386.
(6) Klarer, W.; Urech, E. *Helv. Chim. Acta* **1944**, *27*, 1762.
(7) Lotti, V. J.; Cerino, D.; Kling, P. *J. Auton. Pharmacol.* **1983**, *2*, 169.

0022-2623/83/1826-1769$01.50/0

Table I. Physical Properties of 2-[(Substituted-amino)methyl]-4,5-dihydro-1*H*-imidazoles



compd	R_1	R_2	formula	mp, °C, dec	method	yield, %	recrystn solvent
4a			$C_{17}H_{19}N_3 \cdot HCl$	203.5–206.5	A	70	EtOH–Et_2O
4b	H		$C_{10}H_{13}N_3O \cdot HCl$	180–183	B	44	EtOH–EtOAc
4c		H	$C_{11}H_{15}N_3 \cdot 2HCl$	191–200	C	55	MeOH–EtOH–EtOAc
4d		CH_3	$C_{12}H_{17}N_3 \cdot 2HCl$	249–253	B[a]	67	EtOH–EtOAc
4e		C_2H_5	$C_{13}H_{19}N_3 \cdot HCl$	204–208	B[b]	47	EtOH–Et_2O
4f[c]			$C_{17}H_{25}N_3$	132–134.5	A	37	hexane
4g		H	$C_{10}H_{12}ClN_3 \cdot HCl$	250–254	B[a]	81	EtOH

[a] Refluxed for 24 h, and the precipitated product was filtered. [b] Refluxed for 24 h, and the product was purified by flash chromatography over silica gel with $CHCl_3$ saturated with NH_3. [c] The bis(hydrogen fumarate) salt, mp 143–146 °C dec, was submitted for testing.

renergic antagonist. Antagonist activity is reduced more drastically upon reduction of the phenyl ring of **4a** to cyclohexyl (**4f**). However, even with this structural change, the adrenergic profile is not altered appreciably.

On the other hand, replacement of one of the aromatic rings of phentolamine by hydrogen profoundly changes the adrenergic activities of the parent compound. The phenolic derivative **4b** possess potent α_1- and α_2-adrenergic agonist properties, while the tolyl analogue **4c** is an α_1-agonist with weak α_2-antagonist activities. A phenolic hydroxyl group is not required for α_1/α_2-agonist activity with these monoaryl derivatives, since the 2-chlorophenyl compound **4g** is also a potent α_1/α_2-adrenergic agonist. Other *p*-tolyl derivatives with N-aliphatic substitution (**4d,e**) proved to be mixed α_1-agonists/α_2-antagonists.

It appears from these results that in this 2-(aminomethyl)imidazoline series, arylcyclohexyl or diaryl substitution on nitrogen produces nonselective adrenergic antagonists with potency dependent upon the particular groups used. Although monoaryl derivatives (excluding **4f**) generally appear to be α_1-adrenergic agonists, activity at the α_2 site is more difficult to predict with the examples provided by this limited series. In this case, α_2-agonism or α_2-antagonism is dependent upon the particular nuclear substituent. For example, 3-OH and 2-Cl substituents (**4b,g**) produce α_2-agonists, while the corresponding 4-Me derivative is an α_2-antagonist.

The radioligand binding results for this series of (aminomethyl)imidazolines do not appear to be good indicators of activity in the isolated rat vas deferens. For example, **4c** binds to the clonidine site as well as **4e**, and with higher affinity than **4f**, but in the vas deferens, **4c** is the weakest α_2-antagonist of this series. Also, although **4c–e** are less potent α_1-agonists in the vas deferens than **4b**,**4b** binds to the prazosin site less strongly than **4c–e** or even **4g**, which is a comparable α_1-agonist in the vas deferens.

Experimental Section

Melting points were determined on a Thomas-Hoover capillary melting point apparatus with open capillaries and are uncorrected. ^1H NMR were recorded for all intermediate and final products and are consistent with the assigned structures. Microanalytical results were obtained on all compounds submitted for testing and are within ±0.4% of theoretical values.

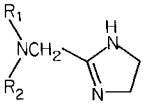
2-[(Substituted-amino)methyl]-4,5-dihydro-1*H*-imidazoles. Method A. 2-[(*N*-Cyclohexyl-4-methylanilino)methyl]-4,5-dihydro-1*H*-imidazole (4f). A mixture of *N*-cyclohexyl-4-methylaniline (3.78 g, 20 mmol) and 2-(chloromethyl)-4,5-dihydro-1*H*-imidazole hydrochloride[6] (1.55 g, 10 mmol) was heated neat at 150 °C for 18 h. After cooling, the solidified product was partitioned between EtOAc and dilute aqueous HCl. The aqueous acid extract was basified, and the crude product was extracted into Et_2O. After the extract was dried (Na_2SO_4) and the Et_2O was removed under reduced pressure, the residue was purified by flash chromatography over silica gel with a 20% MeOH–80% $CHCl_3$ solvent mixture to give 1.0 g of product, mp 128.5–132 °C. An analytical sample, mp 132–134.5 °C, was obtained upon recrystallization from hexane.

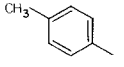
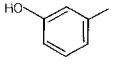
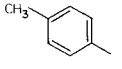
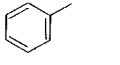
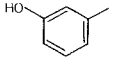
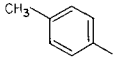
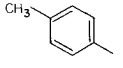
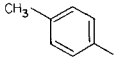
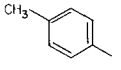
Method B. 2-[(3-Hydroxyanilino)methyl]-4,5-dihydro-1*H*-imidazole Hydrochloride (4b). A solution of 3-hydroxyaniline (1.09 g, 10 mmol) and 2-(chloromethyl)-4,5-dihydro-1*H*-imidazole hydrochloride[6] (775 mg, 5 mmol) in absolute EtOH (10 mL) was stirred at reflux for 2 h. The hot reaction mixture was filtered, and EtOAc was added to the filtrate to precipitate 0.68 g of product, mp 177–180 °C. An analytical sample, mp 180–183 °C dec, was obtained upon recyrstallization from EtOH–EtOAc.

Method C. 2-[(4-Methylanilino)methyl]-4,5-dihydro-1*H*-imidazole Dihydrochloride (4c). A solution of *p*-toluidine (1.07 g, 10 mmol) and 2-(chloromethyl)-4,5-dihydro-1*H*-imidazole hydrochloride[6] (775 mg, 5 mmol) in absolute EtOH (10 mL) was stirred at reflux for 3 h. After the solution was filtered, the filtrate was acidified with excess anhydrous EtOH–HCl. The precipitated solid was redissolved by the addition of MeOH, and the 2HCl salt was reprecipitated with EtOAc to give 720 mg of product, mp 191–200 °C dec.

***N*-Cyclohexyl-4-methylaniline.** A solution of 4-methylaniline (5.4 g, 50 mmol) and cyclohexanone (4.9 g, 50 mmol) in toluene (100 mL) was stirred at reflux under a Dean–Stark H_2O collector for 3 h until removal of H_2O was complete. After the solution was concentrated under reduced pressure, the residue was dissolved in absolute EtOH (150 mL) and hydrogenated over a 5% Pt/C catalyst for 0.5 h until nearly 1 equiv of H_2 had been taken up. The catalyst was removed by filtration, the EtOH was removed under reduced pressure, and the residue was flash chro-

Table II. Radioligand Binding and Rat Vas Deferens Results



compd	R_1	R_2	calf cerebral cortex radioligand binding, K_I, nM		rat vas deferens			
					α_2 activity		α_1 activity	
			clonidine	prazosin	clonidine antagonism: pA_2	agonism:[a] EC_{50}, M	methoxamine antagonism: pA_2	agonism:[a] EC_{50}, M
phentolamine			0.93 ± 0.12	2.1 ± 0.6	7.8 ± 0.0	$>10^{-7}$	8.1 ± 0.1	$>10^{-7}$
4a			1.8 ± 0.2	5.6 ± 1.3	7.5 ± 0.0	$>10^{-6}$	7.6 ± 0.05	$>10^{-6}$
4b	H		7.1 ± 0.9	380 ± 40		7.4×10^{-8} (5.3-14.3)		6.1×10^{-8} (5.8-6.4)
4c		H	14 ± 1	230 ± 20	5.6 ± 0.1	$>2 \times 10^{-6}$		3.4×10^{-7} (3.0-3.9)
4d		CH_3	3.3 ± 0.2	150 ± 40	7.0 ± 0.1	$>2 \times 10^{-6}$		3.0×10^{-7} (2.3-4.1)
4e		C_2H_5	14 ± 1	300 ± 40	7.1 ± 0.0	$>10^{-6}$		1.6×10^{-6} (0.9-3.0)
4f			70 ± 10	424 ± 10	6.5 ± 0.1	$>10^{-6}$	6.0 ± 0.1	$>10^{-6}$
4g		H	1.8 ± 0.1	55 ± 7		5.3×10^{-8} (4.2-6.7)		5.0×10^{-8} (3.6-7.0)

[a] Values in parentheses refer to 95% confidence limits.

matographed on silica gel. Elution with a 30% hexane–70% toluene solvent mixture afforded 5.2 g (55%) of product, mp 41.0–42.0 °C (lit.[8] mp 41 °C).

Radioligand Binding. Assays for the competitive binding of selected compounds to central α-adrenergic binding sites employed radiolabeled clonidine or radiolabeled prazosin, which were obtained from New England Nuclear and Amersham, respectively. [^{3}H]Clonidine (specific activity 22.2–23.8 Ci/mmol) was stored in EtOH–H_2O (7:2) at 0 °C, and [^{3}H]prazosin (specific activity 33 Ci/mmol) was stored in a solution of 1% EtNH in EtOH at 0 °C. The radiochemical purity of these ligands was periodically checked by TLC.

Binding assays were conducted by using frozen sections of calf cerebral cortex (–70 °C). A Brinkmann Polytron PT-10, at setting 6 for 10 s, was used to homogenize the frozen tissue in 20 vol (w/v) of ice-cold 50 nM pH 7.7 Tris-HCl buffer. The resultant homogenate was centrifuged twice at 48000g (Sorvall SS-34 rotor, 2000 rpm, RC-5 centrifuge) for 10 min at 40 °C, with rehomogenization of the intermediate pellet in 20 vol of fresh buffer. This final pellet was resuspended in 50 vol of ice-cold buffer.

Standard displacement assays were run with either 0.20 nM [^{3}H]clonidine or 0.14 nM [^{3}H]prazosin. Triplicate assay tubes contained ^{3}H-labeled ligand, 100 μL of various concentrations of the compound being investigated, 1 mL of tissue homogenate, and 50 mM pH 7.7 Tris-HCl buffer to a final volume of 2 mL. The reaction was initiated by the addition of tissue, and incubation continued for 30 min at 25 °C, at which time it was terminated by rapid filtration through Whatman GF/B glass-fiber filters under vacuum. Each filter was immediately rinsed with 3 × 5 mL aliquots of ice-cold buffer. The filters were removed into 10 mL of PCS (Amersham) and counted on either a Packard Model 2425 or Packard Model 460C scintillation spectrophotometer at approximately 35% efficiency.

Specific binding was defined as the difference between samples with and without 1 μM clonidine or 1 μM prazosin for [^{3}H]clonidine and [^{3}H]prazosin assays, respectively.

Data from binding assays were plotted as log concentration vs. percent inhibition and analyzed by nonlinear least-squares techniques in which 100% maximal inhibition was assumed at high test compound concentrations. The IC_{50} values obtained from such data treatment were used to calculate apparent inhibition constants from eq 1, where [C] is the concentration of

$$K_i = \frac{IC_{50}}{1 + ([C]/K_D)} \quad (1)$$

radioligand employed in the binding assay, and K_D is its receptor dissociation constant (K_D = 0.48 nM for [^{3}H]clonidine and 0.14 nM for [^{3}H]prazosin).

Rat Vas Deferens. Vas deferens were extirpated from Sprague–Dawley rats (250–350 g) and prepared for field stimulation as described previously.[7] Agonist EC_{50} values plus or minus 95% confidence limits were determined by regression analyses of cumulative dose–response curves ($N \geq 2$ tissues) as previously described.[7] α_2-Adrenergic agonist activity was determined by pretreating the tissues with prazosin (78 nM). The α_1- and α_2-adrenergic agonist activity of the test compounds was verified by the ability of prazosin (78 nM) and rauwolscine (280 nM) to completely reverse the contractile enhancement or inhibition, respectively, produced by the test compounds.

Antagonist pA_2 values were estimated on the basis of one or two concentrations of the test compounds using a minimum of three tissues at each concentration.[9] Clonidine and methoxamine were used as α_2- and α_1-adrenergic agonists, respectively, according to the protocols described by Lotti et al.[7]

Acknowledgment. The authors are indebted to Y. Lee and J. Moreau for elemental analyses, P. Bennett, E. Cresson, T. Lyon, and M. Zrada for receptor-binding determinations, J. Murphy for ^{1}H NMR spectra, and D. Cerino and P. Kling for technical assistance with the vas deferens assay. We also thank M. Banker for preparation of the manuscript.

Registry No. **4a**, 87261-69-6; **4a**·HCl, 87261-62-9; **4b**, 87261-70-9; **4b**·HCl, 87261-63-0; **4c**, 87261-71-0; **4c**·2HCl, 87261-64-1; **4d**, 87261-72-1; **4d**·2HCl, 87261-65-2; **4e**, 87261-73-2; **4e**·HCl, 87261-66-3; **4f**, 87261-67-4; **4f** difumarate, 87261-68-5; **4g**, 87261-74-3; **4g**·HCl, 67084-31-5; 2-(chloromethyl)-4,5-dihydro-1*H*-imidazole hydrochloride, 13338-49-3; *N*-cyclohexyl-4-methylaniline, 10386-93-3; 3-hydroxyaniline, 591-27-5; *p*-toluidine, 106-49-0; cyclohexanone, 108-94-1.

(8) Miller, T. G. *J. Org. Chem.* **1966**, *31* 3178.

(9) Mackay, D. *J. Pharm. Pharmacol.* **1978**, *30*, 312.

Parasympatholytic (Anticholinergic) Esters of the Isomeric 2-Tropanols. 2. Non-Glycolates

Edward R. Atkinson,*[†] Donna D. McRitchie-Ticknor,[†] Louis S. Harris,[‡1] Sydney Archer,[‡2] Mario D. Aceto,[‡1] J. Pearl,[‡] and F. P. Luduena[‡]

Arthur D. Little, Inc., Cambridge, Massachusetts 02140, and Sterling-Winthrop Research Institute, Rensselaer, New York 12144. *Received March 31, 1983*

The 19 esters in Table I were prepared from (+)-2α-tropanol, (–)-2β-tropanol, (±)-3-quinuclidinol, and a variety of non-glycolic acids in order to compare their central and peripheral activities with those of the glycolates reported in the previous paper. The results (Table II) showed that esters **6** and **17** were approximately equivalent to one another and to atropine, that **8** was equal in both central and peripheral activity to reference glycolates, that **9** and **19** were less active than **8** but **9** had a substantially reduced central activity, and that **10** and **11** were more active than the methoxy analogue reported earlier.

All the natural and many of the synthetic anticholinergic drugs (such as methixene and methantheline) are not glycolates. Some of these (such as scopolamine) have potent CNS activity. It was shown previously[3] that some glycolate esters of the 2-tropanol isomers **1** and **2** have powerful CNS effects. It was therefore of interest to de-

† Arthur D. Little, Inc.
‡ Sterling-Winthrop.
* Corresponding address: 163 Gray St., Amherst, MA 01002.

(1) Present address: Department of Pharmacology, Medical College of Virginia, Richmond, VA 23298.
(2) Present address: Department of Chemistry, Rensselaer Polytechnic Institute, Troy, NY 12181.
(3) Atkinson, E. R.; McRitchie, D. D.; Shoer, L. F.; Harris, L. S.; Archer, S.; Aceto, M. D.; Pearl, J.; Luduena, F. P. *J. Med. Chem.* **1977**, *20*, 1612.

0022-2623/83/1826-1772$01.50/0